Clonal propagation of guava (*Psidium guajava* Linn.) by stem cutting from mature stockplants

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Abstract: The study describes the scope of clonal propagation of guava (*Psidium guajava* Linn.) by stem cutting collected from mature stockplants. Cuttings were treated with 0, 0.2%, 0.4% and 0.8% IBA solution and rooted in the non-mist propagator. Rooted cuttings were allowed to grow in the polybags filled with soil and cow-dung mixed in the ratio of 3: 1 (by volume) for three months to assess the steckling capacity and initial growth performance. The study reveals that the species is amenable for clonal propagation by mature stem cutting. The highest rooting percentage (60%) was observed in the cuttings treated with 0.4% IBA solution followed by 0.2% IBA and the lowest was in controlled cuttings. The maximum number of primary root (32.7) was developed in the cuttings treated with 0.8% IBA solution followed by 0.4% IBA and the lowest was in the cuttings without IBA treatment. The highest survival percentage (70.9) was observed in the cuttings rooted with 0.4% IBA treatment and the lowest (58.3) was in the cuttings without any treatment. However, there was no significant variation in height growth of cutlings due to IBA treatments in rooting.

Key words: *Psidium guajava* Linn.; Non-mist propagator; Stockplants; Stem cutting; Rooting ability; Stecklings

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Introduction

Guava (*Psidium guajava* Linn.), belonging to the Family Myrtaceae, is originated in the tropical South America (Hayes 1970; Pathak and Ojha 1993) and grows wild in Bangladesh, India, Thailand, Brazil, Florida, West Indies, California and also in several other countries (Bailey 1960). Bangladesh is one of the major guava producing countries of the world. It is grown through out the country in home gardens and in Agroforestry production systems. About nine thousand hectares of lands in the country are under guava cultivation, yielding a total of four thousand metric tons per year (Razzaque *et al*. 2000).

Guava is very popular fruit in Bangladesh to the people of all ages in all classes of the society for its good taste, nutritional value and lower price (Rahman *et al*. 2003). It plays a vital role in fulfilling the vitamin C deficiency among the people of the country (BBS 2000) since 100 g of fruit contains about 260 mg of vitamin C (Pandis 1970; FAO 1984; Rahman *et al*. 2003), which is 2–5 times higher than the fresh orange. The barks and leaves of the trees posses some medicinal values. Roots and young leaves are astringent, extremely useful in strengthening the stomach. The wood is hard and tough (Drurey 1985), used as posts for rural house buildings. Tannin and dye can be made from the leaves (Siddiki and Ali 1994).

Guava plants can successfully be propagated by both sexual (by seed) and asexual methods (by cutting, layering, budding and grafting) of regeneration. It is usually propagated by the method of freshly extracted seeds and air layering (Singh, 1985). However, the number of plant obtained by the methods of layering is inadequate (Singh 1986). Seed originated guava plants cannot maintain the genetic purity of the variety due to the segregation and recombination of characters during sexual reproduction. In the contrary, clonal propagation of guava can be considered to avoid the segregation of genetic variety and maintain the quality of fruits. There are several reports, which have examined the possibilities of propagation of guava by using stem cutting from seedling-originated juvenile stockplants in which genetic segregation is possible. Propagation by cuttings from mature trees may be one of the important options to avoid the genetic segregation and maintain the quality of the variety. However, the information regarding the rooting ability of the cutting obtained from mature stockplants of the species is very scarce. The present study was therefore designed to examine the possibilities of propagation of guava through stem cutting collected from mature stockplants.

Materials and methods

The study was conducted over a period of six month from April to September 2004 in the nursery of Institute of Forestry and Environmental Sciences at Chittagong University campus, Bangladesh (22°27′ N, 91°47′ E). Cuttings were collected from the superior phenotype trees grown in the study area. Branches of the stockplants were trimmed for shoot production at the beginning of the study. The sprouted shoots were collected from the trimmed branches and soaked in water immediately.

One node cuttings with two leaves trimmed to half were made for the rooting trial. Length and diameter of cuttings was kept indifferent among the treatments to avoid the non-treatment variations. The average height and diameter of the cuttings was 4.2 cm to 4.8 cm and 2.8 mm to 3.0 mm, respectively. Cuttings were then immersed briefly in a solution of fungicide, Diathane M45 (Rohm & Co. Ltd., France; 2 g. per litre of water) to avoid fungal infection and kept under shade for 10 min in open air. The cuttings were then treated with 0, 0.2%, 0.4% and 0.8% IBA (Indole 3-butyric acid) solution to explore the rooting ability. For IBA treatment base of the cuttings was briefly dipped into the solution for 15 s. Treated cuttings were planted into perforated plastic trays (12 cm depth) filled with coarse sand mixed with

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fine gravel and placed into a non-mist propagator (Fig. 1) for rooting in a completely randomized block design (Kamaluddin 1996).

A total of 180 cuttings were placed under four different treatments with three replications in a non-mist propagator. Cuttings were planted in 12 trays, 3 trays for each treatment (0, 0.2%, 0.4% and 0.8% IBA solution) and each tray (containing 15 cuttings) served as a plot. Thus the number of replicate cuttings per treatment was 45. The cuttings were watered once only just after setting into the propagator in every case. A light spray was done every morning with a hand spray till the transfer of rooted cuttings from the propagator.

The cuttings in the propagator rooted four weeks after setting into the propagator. The cuttings were subjected to weaning before transfer them into polybags, particularly towards the end of rooting period during root lignifications. For weaning, the propagator was kept open at night for three days and then at day and night for another three days. Weaning is usually done to harden the rooted cuttings in prevailing adverse environment outside the propagator. It increases the steckling capacity of the rooted cuttings. After weaning, all rooted cuttings were transferred into polybags filled with soil and decomposed cow-dung at a ratio of 3:1 (by volume). Number of primary roots developed in each cutting was recorded before planting the rooted cuttings into the polybags. The rooted cuttings were then allowed to grow for three months in open sun. Finally the survival percentage and the initial height growth of each cutlings were recorded three months after transferring them into the polybags.

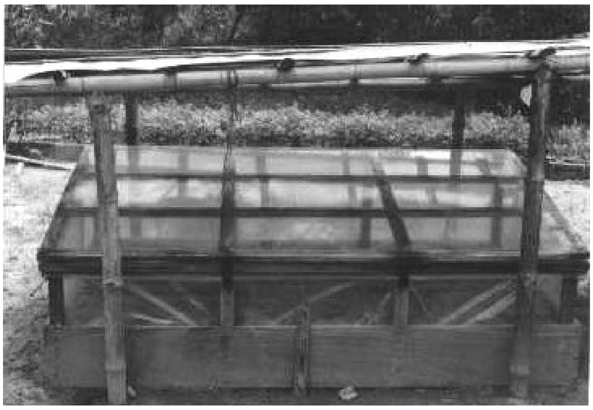

Fig. 1 A non-mist propagator1.8 m in length and 1 m in breath with a height of 60 cm on one side and 45 cm on the other side

All data were analyzed by the software of Microsoft Excel, SPSS. Possible treatment variations were explored by the results of Analysis of Variance (ANOVA) and Duncan's Multiple Range Test (DMRT).

Results and discussion

Rooting ability of cuttings

Rooting percentage

Cuttings of guava obtained from the mature stockplants rooted well in the experiments. Rooting percentage of cuttings varied from 41.7 to 60.0 among the cutting types (Fig. 2). The highest rooting percentage (60%) was observed in the cuttings treated with 0.4% IBA solution followed by 0.2% IBA and the lowest (41.7%) was in the cuttings without IBA treatment.

Rooting percentage of the cuttings was significantly increased with the IBA treatment in the present study. The result was not possible to compare with the study of the other authors due to lacking of related references, which have discussed the rooting ability of the mature stem cutting of guava. However, Hossain *et al.* (2004) mentioned that the rooting percentage of juvenile cuttings of guava varied from 93.3 to 100 without IBA treatment. Again, in a separate experiment Hossain and Kamaluddin (2005) reported that the rooting percentage was 100 in the cuttings obtained from juvenile stockplants (seedlings) of guava. Rooting percentage in the present study was much lower compared with that of Hossain *et al.* (2004) and Hossain and Kamaluddin (2005). This might be due to the hardwood cuttings of the mature stockplants of guava since Kilany and Gabr (1986) reported that the rooting in hardwood cuttings was very poor (1.64%–4.67% only); it ranged from 18.3% to 57.5% in leaf bud cuttings and was highest (81.4%) in semi-hardwood cuttings treated with 0.25% IBA + 0.01% alpha-napthol. Similarly, softwood cuttings generally rooted better (the highest rooting percentage, 70) than the semi-hardwood cuttings with treatment (2 nodes + 4 leaves) as reported by Pereira *et al.* (1983).

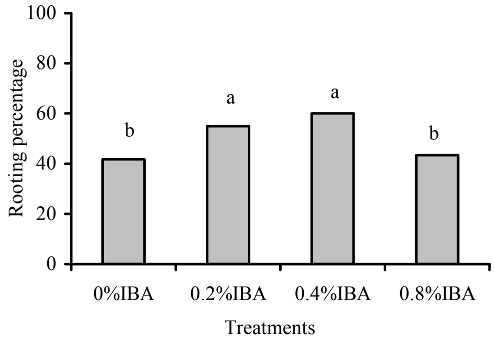

Fig. 2 Rooting percentage of guava cuttings under various treatments. The same letters indicate no significant difference at $p<.05$ (ANOVA and DMRT).

Again, the method to enhance rooting percentage of stem cuttings was reported by many authors. For instance, Al-Obeed (2000) reported that the maximum rooting percentage (62.9%) obtained in guava cuttings treated with IBA + catechol at 0.1%, followed by cuttings treated with NAA (Naphthalene acetic acid) + Catechol at 0.1% (59.6%) and the lowest rooting percentage (19.8%) was in the untreated cuttings. Hossain *et al.* (2002) reported that the exogenous auxin (0.4% IBA) significantly ($p<0.05$) enhanced the rooting percentage of cuttings of jackfruit (*Artocarpus heterophyllus* Lam.). Similar result was also reported by Abdullah *et al.* (2005) and mentioned that the highest rooting percentage of *Baccaurea sapida* Muell. Arg mature stem cutting was 65 in 0.4% IBA treatment and the lowest (15%) was in controlled cuttings.

Root number of cuttings

The number of primary root developed per cutting was significantly affected by the IBA treatment in guava cuttings. Root number was increased gradually with the increment of the IBA concentration. The highest number of root per cutting (32.7) was observed in the cuttings treated with 0.8% IBA solution and the

lowest (8.7) was in cuttings without IBA treatment (Figs. 3 and 4).

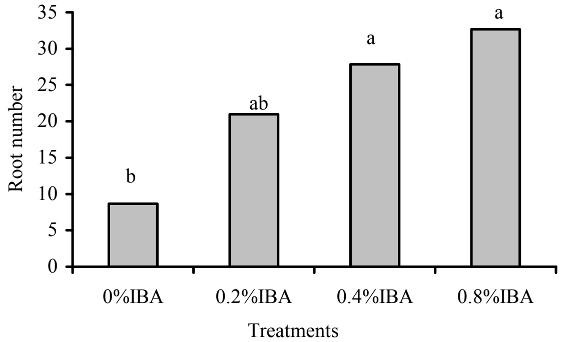

Fig. 3 Root number of guava cuttings under various treatments. The same letters indicate no significant difference at p<0.05 (ANOVA and DMRT).

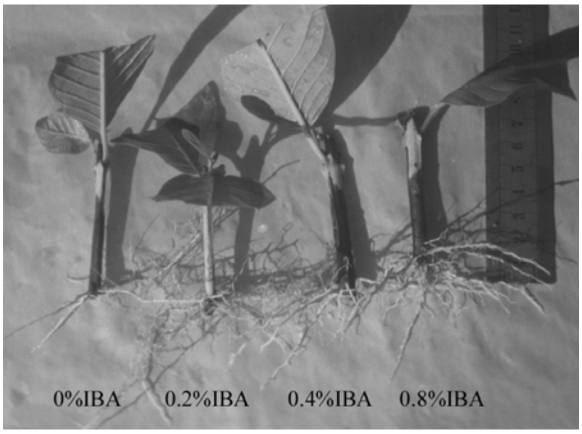

Fig. 4 Rooting ability of guava stem cuttings from mature stockplant under various treatments.

The result of the present study regarding the average number of primary root produced in the cuttings was supported by many authors. Al-Obeed (2000) mentioned that the cuttings of guava treated with IBA in combination with catechol at 0.05% and 0.1% gave the highest number of roots (31.1) while the control cuttings produced only 9.1 roots per cutting. Sen (2006) mentioned that the average number of root of *Flacourtia jangomas* cuttings was significantly enhanced due to the applied IBA. The highest number of root was developed in 0.4% IBA treated cuttings and the lowest (2.1) was in cuttings without treatment. Hossain *et al.* (2002) mentioned that exogenous auxin (0.4% IBA) significantly (p <0.05) increased the root number of cuttings of jackfruit. In a separate experiment Hossain *et al*. (2004) recorded the highest number of root per cutting (16.9) of *Chickrassia velutina* treated with 0.4% IBA solution compared with 6.3 in cuttings without IBA treatment. Abdullah *et al*. (2005) reported the highest number of root in *B. sapida* Muell. Arg cuttings treated with 0.8% IBA solution. Dias *et al.* (1999) reported the highest number of root developed in the cuttings of *Platanus acerifolia* treated with 0.6% IBA 120 days after setting the cuttings in the greenhouse. Again, Kamaluddin *et al.* (1998) reported that applied auxin significantly increased rooting ability of *C. velutina* cuttings in non-mist propagator.

Applied rooting hormone enhanced the rooting ability of cuttings was reported by many authors including Hossain (1999), Hossain *et al.* (2002), Hossain *et al.* (2004), Abdullah *et al.* (2005), Sen (2006), Dias *et al.* (1999), Kamaluddin (1996), Kamaluddin and Ali (1996) and Kamaluddin *et al.* (1998). Applied IBA treatment is believed to activate polysaccharide hydrolysis, and as a result, the contents of physiologically active sugar increased providing materials and energy for merestamatic tissues and later for root primordia and roots. Hassig (1983) examined the function of endogenous root forming components of plants, which had auxin component and non- auxin components. He demonstrated that auxin component was required for development of callus in which root primordia initiated, but for subsequent primordia development both auxin and non-auxin components were needed. It may be possible that in cuttings with optimum amount of endogenous auxin content and increasing of root number reflected the effect of applied auxin.

Steckling capacity of rooted cuttings

Survival percentage

Applied auxin significantly enhanced the survival percentage of guava cutlings (Table 1). The survival percentage varied from 70.9 to 58.3 across the treatments. The highest survival percentage (70.9%) was observed in the cuttings rooted with 0.4% IBA treatment followed by 68.3% in the 0.2% IBA and the lowest (58.3%) was in cuttings rooted without IBA treatments.

Similar result was reported by Sen (2006) and mentioned that the best survival percentage of the *F. jangomas* cutlings (85%) was observed when rooted with 0.4% IBA treatment followed by 0.2% IBA and the lowest (40%) was in the cuttings without treatment. Again, Nath and Barooah (1992) mentioned that the survival percentage of the rooted cuttings of *F. jangomas* was significantly higher in the cuttings treated with 2500, 3000 or 0.35% IBA than those in controlled cuttings. In the same token, Kamaluddin (1996) reported the highest survival percentage of the cutlings of *E. camaldulensis* and *Acacia* hybrid rooted with 0.4% IBA solution. The reasons for increasing the survival percentage in the cutlings rooted with IBA treatment was not possible to explain due to lack of related references. However, the cutlings of some species like *Vitex negundo* showed better survival in the cuttings rooted without IBA treatment than the cuttings rooted with IBA (Afroz 2006). The reason for the higher or lower survival in the cutlings of many species treated with IBA was not clear to us. This might be primarily due to the inherent capacity of the species to be rooted and survived.

Table 1. Survival percentage and initial height growth of guava cutlings under various treatments

Source of variation	Treatments				p
	0%IBA	0.2%IBA	0.4%IBA	0.8%IBA	
Survival percentage	58.33^b	68.33^a	70.96^a	68.22^a	**
Height (cm)	19.65^a	19.39^a	22.59^a	20.21^a	NS

** indicates the significant difference at p<0.1 and NS indicates no significant difference at p<0.05. The same superscript letters indicate no significant difference at p<0.05 (ANOVA and DMRT).

Height increment of cuttings

The height growth of cutlings was not significantly affected by IBA treatments (Table 1). The height growth was varied from 19.4 cm to 22.6 cm among the treatments three months after

transplanting the rooted cuttings into the polybags. The maximum height growth was in cutlings rooted with 0.4% IBA treatment. However, there was no study found regarding the effect of IBA treatment on height growth of the cutlings to compare the result of the present study.

Conclusion

Guava is one of the most important fruit species in Bangladesh, which deserve vegetative propagation for many reasons. Clonal propagation through stem cutting might be one of the effective methods of vegetative propagation for the species since it rooted well in the cuttings collected even from the mature stockplants. The highest rooting percentage of guava cuttings in the present study was observed in the cuttings treated with 0.4% IBA solution while the maximum number of root was in cuttings treated with 0.8% IBA solution, which was remarkably responsive to the applied auxin (IBA). However, the highest survival of the cuttings was recorded in the cuttings rooted with 0.4% IBA solution. Therefore,, considering the rooting percentage, root number in the cuttings and their steckling capacity under different treatments, clonal propagation by mature stem cutting with 0.4% IBA treatment may be recommended for the species. However, the genetic constituents in the propagules produced in the present study and their field performance could not be determined due to the limitation of time. This might be one of the important aspects of future study.

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